ESC HEART FAILURE
ESC Heart Failure 2015; **2**: 121–132
Published online in Wiley Online Library (wileyonlinelibrary.com) **DOI:** 10.1002/ehf2.12049

ORIGINAL RESEARCH ARTICLE

Heterogeneous responses of systolic and diastolic left ventricular function to exercise in patients with heart failure and preserved ejection fraction

Mario Kasner[1]*, David Sinning[1], Jil Lober[1], Heiner Post[2], Alan G. Fraser[3], Burkert Pieske[2,4], Daniel Burkhoff[5] and Carsten Tschöpe[2,4,6]

[1]*Department of Cardiology, Charité-Universitätsmedizin Berlin, Campus Benjamin Franklin, Hindenburgdamm 30, Berlin, Germany;* [2]*Department of Cardiology, Charité-Universitätsmedizin Berlin, Campus Virchow, Berlin, Germany;* [3]*Wales Heart Research Institute, Cardiff University, Cardiff, UK;* [4]*Germany Centre for Cardiovascular Research (DZHK), Berlin, Germany;* [5]*Division of Cardiology, Columbia University, New York, NY, USA;* [6]*Berlin-Brandenburg Center for Regenerative Therapies, Charité-Universitätsmedizin Berlin, Campus Virchow, Berlin, Germany*

Abstract

Aims This study aimed to evaluate ventricular diastolic properties using three-dimensional echocardiography and tissue Doppler imaging at rest and during exercise in heart failure with preserved ejection fraction (HFpEF) patients with borderline evidence of diastolic dysfunction at rest.

Methods and results Results obtained from 52 HFpEF patients (left ventricular ejection fraction $\geq$50%) identified on the basis of heart failure symptoms and *E*/*E*′ values between 8 and 15 were compared with those obtained in 26 control patients with no evidence of cardiovascular disease. Mitral flow patterns, tissue Doppler imaging, and volume analysis obtained by three-dimensional echocardiography were performed at rest and during bicycle exercise. Diastolic compliance was indexed by the *E*/*E*′ ratio and left ventricular end-diastolic volume [(*E*/*E*′)/EDV]. There were no significant differences in end-diastolic volume (EDV), stroke volume (SV), or ejection fraction at rest between groups. In 27 of the 52 patients, *E*/*E*′ increased during exercise (11.2 ± 3.7 to 16.8 ± 10.5), driven by a failure to augment early diastole (*E*′). This correlated with a fall in SV and was associated with an increase in the diastolic index (*E*/*E*′)/EDV as a measure for LV stiffness (0.122 ± 0.038 to 0.217 ± 0.14/mL), indicating that impaired diastolic reserve (designated PEF-I_DR) contributed to exercise intolerance. Of the 52 patients, 25 showed no changes in *E*/*E*′ during exercise associated with a significant rise in SV and cardiac output, still inappropriate compared with controls. Despite disturbed early diastole (*E*′), a blunted increase in estimated systolic LV elastance indicated that impaired systolic reserve and chronotropic incompetence rather than primarily diastolic disturbances contributed to exercise intolerance in this group (designated PEF).

Conclusion Three-dimensional stress echocardiography may allow non-invasive analysis of changes in cardiac output that can differentiate HFpEF patients with an inappropriate increase or a fall in SV during exercise. Impaired systolic or diastolic reserve can contribute to these haemodynamic abnormalities, which may arise from different underlying pathophysiologic mechanisms.

Keywords Diastole; Heart Failure; 3D Echocardiography; Exercise; Haemodynamics

Received: 26 April 2015; Revised: 10 June 2015; Accepted: 17 June 2015
**Correspondence to: Mario Kasner, Department of Cardiology, Charité-Universitätsmedizin Berlin, Campus Benjamin Franklin, Hindenburgdamm 30, 12200 Berlin, Germany. Tel: +49 30 8445-2383, Fax: +49 30 8445-4648. Email: mario.kasner@charite.de*

Introduction

Heart failure with preserved ejection fraction (HFpEF) is a common clinical syndrome with high morbidity and mortality that is increasing in prevalence with the ageing population.[1,2] The underlying pathophysiological mechanisms leading to the clinical symptomatology in patients with HFpEF are still incompletely understood but are believed to include non-cardiac and cardiac components.[3–13] In addition to factors such as abnormal ventricular–arterial coupling and reduced

myocardial contractility, decreased ventricular compliance causing abnormal diastolic filling is considered to be important in explaining pulmonary congestion.[14–17] However, the heterogeneity of the disease and the lack of fundamental understanding of the underlying pathophysiologic mechanisms have hindered progress in developing therapies and even lead to differences of opinion over how to define and diagnose patients with HFpEF. Indeed, recognition that the HFpEF population includes patients with an extremely diverse set of underlying pathophysiologies has only recently been introduced to target recruitment of relevant subgroups into clinical studies investigating therapies aimed at specific underlying mechanisms of the disease.[18,19]

In clinical practice, these patients are diagnosed by criteria established by the European Society of Cardiology (ESC).[20] However, it can be difficult to establish a diagnosis in patients who are asymptomatic at rest but suffer from exercise intolerance despite preserved ejection fraction (EF) without evidence of left ventricular (LV) hypertrophy or left atrial (LA) dilatation and with borderline echocardiographic Doppler diastolic filling parameters (e.g. *E*/*E*' between 8 and 15). The presence and nature of cardiac dysfunction or other non-cardiac contributing factors in such patients often remain unrecognized. In such cases, haemodynamic and echocardiographic assessments during exercise can provide significant insights into mechanisms,[21] in particular through assessment of changes in heart rate (HR), stroke volume (SV), cardiac output (CO), and pressure–volume relationships, that might be manifested only during exercise. However, this approach is not included into the ESC guidelines.

Therefore, we investigated non-invasively derived parameters of cardiovascular properties to provide insight into the physiology of exercise-induced symptoms among patients meeting conventional criteria for HFpEF and with borderline evidence of diastolic dysfunction by Doppler echocardiography at rest. We used three-dimensional (3D) echocardiography (3DE) for accurate assessment of LV volumes and changes in volumes during exercise, which, in turn, allows for assessment of changes in diastolic properties via pressure–volume analysis at rest and during exercise.

Methods

Patient population

Patients who presented with heart failure symptoms during exercise despite normal EF (≥50%) and characterized as having 'borderline' LV diastolic dysfunction as indexed by an *E*/*E*' ratio between 8 and 15 were considered eligible for this study. All patients had at least one episode of heart failure-related hospitalization in the past year, suffering from dyspnoea, orthopnoea, or paroxysmal nocturnal dyspnoea. All eligible patients were carefully screened for non-cardiac causes of heart failure symptoms; patients with significant lung or renal disease or overt volume overload were excluded. Patients with atrial fibrillation, valvular disease more than mild, significant coronary artery disease, and/or hypertrophic cardiomyopathy were excluded by means of electrocardiography, laboratory values, and/or echocardiography. None of the patients had a history of acute coronary syndrome or significant obstruction of any coronary vessel, and none had prior coronary stenting. Severity of heart failure symptoms was quantified with the New York Heart Association (NYHA) classification. Exercise tolerance was assessed by bicycle ergometry. N-terminal pro-brain natriuretic peptide (NT-proBNP) plasma levels were also obtained at baseline (Elecsys 2010, Roche Diagnostics GmbH, Mannheim, Germany).[22] A total of 52 patients met the inclusion criteria and were enrolled in this study as the HFpEF group.

A control group consisted of 26 healthy volunteers recruited from our outpatient clinic who were presented for preventive examination. Control subjects had normal systolic and diastolic LV functions by standard echocardiographic criteria, and they underwent the same exercise protocol as the HFpEF group. Cardiac conditions were stable before testing in all participants, and all medications were withheld for 72 h prior to examination. All participants provided informed written consent.

Bicycle exercise protocol

Transthoracic echocardiography (VIVID System Seven Dimension or Vivid E9, GE Ultrasound, Horten, Norway) was performed using 2.5 MHz transducer probes for two-dimensional (2D) acquisition and 3 V probe for multiplane and 3D full-volume acquisitions continuously throughout a bicycle exercise in a semi-supine position using a dedicated bed. Exercise started with a 25 W load, which was increased every 2 min by 25 W until reaching a maximal predicted workload and/or reaching maximal predicted HR (220-age). Echocardiographic images were acquired *at rest* before starting exercise, at *low-level* exercise (at ~30% of maximal predicted workload) and at *high level* (at >65% of maximal predicted workload).[23] Patients continued the exercise test after the high level until maximal workload or maximal HR was achieved. Blood pressure and 12-lead echocardiography were recorded for each exercise level.

Doppler and three-dimensional echocardiography

The assessment of diastolic function included pulsed-wave Doppler measurements of the early (*E*) and late (*A*) mitral inflow velocities, deceleration time of early LV filling, and

the peak early (*E*′) and late (*A*′) diastolic velocities of the septal and lateral mitral annulus by tissue Doppler in the four-chamber view over at least three cardiac cycles. Accordingly, the ratio of early to late annular velocity (*E*′/*A*′) and LV filling index defined as the transmitral flow velocity-to-annular velocity ratio (*E*/*E*′) were determined for septal and lateral walls, as recommended.[20] In case of mitral inflow wave fusion (*E* and *A*) due to higher HR, the ratio of fusion *E*–*A* wave and *E*′ was used. Changes in LV volume were obtained using full-volume assessment over four cardiac cycles during breath hold. Patients with low window quality and requirements needed for the reliable full-volume analysis were not included (four HFpEF and one control). A commercially available software package (TomTec Imaging Systems 4D LV-Function 2.2.1™, Unterschleißheim, Germany) was used for post-acquisition volume analysis at an EchoPAC PC Workstation (GE Vingmed Ultrasound AS, Horten, Norway), which provided semi-automatic measurements of LVEDV, end-systolic volume (ESV), SV, CO, EF, and systolic dyssynchrony index (SDI).[24]

Chamber sizes were evaluated using standard procedures, including LV mass index and LA volume index. LA volume was measured according to the biplane area–length method in four-chamber and two-chamber views and was indexed to body surface area. Cardiac cycles were recorded in a cine loop format. Images were stored digitally for subsequent offline analysis. Interpretation of the echocardiograms was performed by two independent investigators blinded to the results of the other. To determine interobserver variability, 10 patients were randomly selected and independently assessed by another echocardiographer blinded to patient data and previous results.

Haemodynamic parameters

E/*E*′ was taken as a non-invasive means of estimating LV end-diastolic pressure. Simultaneous assessment of a 3D volume allowed estimation of diastolic stiffness as the ratio between *E*/*E*′ and LVEDV: (*E*/*E*′)/EDV. We validated this non-invasive estimation of LV stiffness recently[25] in a subset of patients in whom LV 3D volumes and *E*/*E*′ were measured simultaneously with invasive measurement of pressure–volume relationships by the conductance catheter method. Briefly, we directly measured the end-diastolic pressure–volume relationship during transient preload reduction by vena cava balloon occlusion. Data were then fitted to the exponential curve: $\mathrm{LVEDP} = c * \exp(\beta * \mathrm{LVEDV})$, where β is LV stiffness. The non-invasive estimates of stiffness [(*E*/*E*′)/EDV] correlated with the invasive measurements (β, $r = 0.85$, $P < 0.001$). Using simultaneous blood pressure measurements, end-systolic pressure (ESP) was estimated, ESP = 0.9 × systolic blood pressure,[26] and accordingly, an estimate of end-systolic stiffness (E_{LV}) could be obtained by $E_{LV} = \mathrm{ESP}/\mathrm{ESV}$.[27,28] Arterial stiffness (Ea) as a measure of net arterial load was approximated by the ratio of ESP and SV.[29]

Longitudinal 2D strain measurements in apical four-chamber and two-chamber views were performed at rest and during exercise using speckle tracking method (greyscale) at frame rates over 60 bps, preferably a minimum rate of HR/2. A standard software package available on the GE ECHOPAC station was used for this analysis as described previously.[30]

Statistical analysis

SPSS software (version 15.0, SPSS Inc, Chicago, IL, USA) was used for statistical analysis. Continuous variables were expressed as mean values with standard deviations. Two-sample comparisons between HFpEF patients and controls were performed using *t*-test if variables were normally distributed, the Mann–Whitney U-test for non-normally distributed data, and the χ^2 test for categorical data. Correlation analysis was provided using Pearson's correlation coefficients for normally distributed continuous data, and Spearman coefficient was used for non-normally or non-continuous data. Regression analysis was performed to determine the exact relations between (*E*/*E*′)/EDV and SV values. Multivariate regression analysis was used to examine if (*E*/*E*′)/EDV was an independent predictor for cardiac performance. A *P*-value less than 0.05 was considered statistically significant in all analyses. The authors had full access to the data and assume responsibility for its integrity. All authors have read and approved the manuscript as written.

Results

Patient characteristics

Patient characteristics are summarized in *Table 1*. There were no significant differences between HFpEF patients and control subjects with respect to age, gender, race, and body mass index. HFpEF patients had higher systolic blood pressures (consistent with high prevalence of hypertension). Most of the HFpEF patients were classified as NYHA II, and accordingly, NT-proBNP levels were significantly increased in HFpEF group compared with control subjects. However, 16% of these patients had NT-proBNP levels in the normal range. HFpEF patients showed impaired exercise tolerance on the bicycle exercise test compared with the controls. There was a high prevalence of co-morbid diseases in the HFpEF group and, accordingly, a relatively high rate of use of cardiovascular medications.

Table 1 Patient characteristics (variable expressed as mean ± standard deviation)

	Controls ($n = 26$)	HFpEF ($n = 52$)	P
Demographics			
Gender m/f, *n*	13/13	27/25	0.532
Age, years	48 ± 11	55 ± 12	0.055
BMI, kg/m^2	24.6 ± 4.3	27.3 ± 5.0	0.051
Waist, cm (m/f)	95 ± 13/ 88 ± 19	106 ± 14/ 90 ± 12	0.036/ 0.812
BP systolic, mmHg	126 ± 17	141 ± 22	0.041
BP diastolic, mmHg	70 ± 9	77 ± 14	0.084
NYHA II/III, *n* (%)	0 (0)/0 (0)	39(75)/9 (17)	—
NT-proBNP, pg/mL	65 ± 45	348 ± 716	0.020
Exercise capacity, W	178 ± 61	102 ± 35	<0.001
Concomitant disease, *n* (%)			
Arterial hypertension	0 (0)	28 (54)	—
Diabetes mellitus	0 (0)	10 (19)	—
Obesity (BMI > 30 kg/m^2)	4 (15)	15 (29)	0.115
Hyperlipoproteinaemia	5 (19)	23 (44)	0.093
Smoking	5 (19)	17 (32)	0.142
Medications, *n* (%)			
Beta-blocker	0 (0)	22 (42)	—
ACEI/ARB	0 (0)	25 (48)	—
Diuretics	0 (0)	23 (46)	—
Calcium channel blocker	0 (0)	13 (25)	—

ACEI, angiotensin-converting enzyme inhibitor; ARB, angiotensin 1 receptor blocker; BMI, body mass index; BP, blood pressure; HFpEF, heart failure with preserved ejection fraction; NT-proBNP, N-terminal pro-brain natriuretic peptide; NYHA, New York Heart Association class.

Baseline echocardiography

The results of baseline echocardiographic evaluations are summarized in *Table 2*. All of the HFpEF patients had cardiac dimensions within normal limits. LV mass index and LV mass–volume ratio (a sign of concentric hypertrophy) were borderline elevated in HFpEF patients. Patients with HFpEF had only mildly increased atrial size compared with controls.[31] Resting 3DE LV volumes are also summarized in *Table 2*. There were no significant differences in LVEDV, SV, CO, or LVEF at rest between the groups.

Diastolic function at rest and during exercise

Mitral flow patterns at rest (*Table 2*) showed significantly decreased *E*/*A* ratio with elevated deceleration time of early mitral flow and isovolumetric relaxation time in HFpEF patients. All HFpEF patients showed only mild diastolic dysfunction at rest. Tissue Doppler imaging (TDI) revealed decreased *E*′/*A*′ ratio and confirmed elevated *E*/*E*′ (indexes of LV filling dynamics) in the HFpEF group.

In control patients, there were no significant changes in *E*/*E*′, as both *E* and *E*′ increased simultaneously during exercise (*Table 3*). In contrast, HFpEF patients responded

Table 2 Heart dimensions, mitral flow, and tissue Doppler imaging in patients with HFpEF compared with controls (variable expressed as mean ± standard deviation)

	Controls ($n = 26$)	HFpEF ($n = 52$)	P
Heart dimensions			
LA, mm	34 ± 4	37 ± 6	0.080
LAVI, mL/m^2	19 ± 12	26 ± 15	0.057
Septum, mm	10 ± 1.5	12 ± 2.1	0.007
Posterior wall, mm	10 ± 1.5	11 ± 1.6	0.017
LVEDD, mm	49 ± 5.1	47 ± 5.2	0.231
LVMI, g/m^2 (m/f)	99 ± 20/ 79 ± 19	117 ± 31/ 93 ± 23	0.168/ 0.088
LVMV, g/mL	1.7 ± 0.5	2.0 ± 0.6	0.045
LVEDVI, mL/m^2	57 ± 12	56 ± 15	0.528
LVESVI, mL/m^2	22 ± 6	23 ± 7	0.662
SV, mL	68 ± 18	63 ± 21	0.288
EF, %	62 ± 5	61 ± 7	0.261
Mitral flow			
E, cm/s	74 ± 15	75 ± 15	0.951
A, cm/s	62 ± 17	77 ± 18	0.001
E/*A*	1.25 ± 0.39	0.98 ± 0.30	0.001
DT, ms	186 ± 27	209 ± 44	0.086
IVRT, ms	87 ± 13	100 ± 13	0.028
Tissue Doppler			
S'_{mean}, cm/s	8.4 ± 1.7	7.7 ± 2.0	0.108
E'_{mean}, cm/s	13.1 ± 3.1	7.4 ± 2.1	<0.001
A'_{mean}, cm/s	8.5 ± 2.1	8.0 ± 2.3	0.359
E' / A'_{mean}	1.64 ± 0.52	1.0 ± 0.47	<0.001
E' / E'_{mean}	5.9 ± 1.2	11 ± 3.0	<0.001

DT, deceleration time of early mitral flow; *E*/*A*, the ratio of early (*E*) to late (*A*) mitral flow peak velocities; *E*′/*A*′, ratio of early to late annular velocity; *E*/*E*′, LV filling index; EF, ejection fraction; HFpEF, heart failure with preserved ejection fraction; IVRT, isovolumetric relaxation time; LA, left atrial diameter; LAVI, left atrial volume index; LVEDD, LV end-diastolic diameter; LVEDVI, LV end-diastolic volume index; LVMI, LV mass index; LVESVI, LV end-systolic volume index; *S*′, *E*′, and *A*′, systolic, early, and late diastolic peak velocities of mitral annulus at lateral site, respectively; SV, stroke volume.

in one of two ways based on changes in *E*/*E*′ driven by a blunted increase of *E*′. One group (designated PEF), composed of 25 of the 52 patients (48%), did not show changes in *E*/*E*′ (10.5 ± 2.4 to 9.9 ± 2.7) or (*E*/*E*′)/EDV (0.099 ± 0.028 to 0.087 ± 0.031/mL), suggesting impaired systolic rather than diastolic reserve (*Figure 1* and *Table 3*), although very early diastolic dysfunction may be also disturbed with lower *E*′ increment during exercise compared with controls (*Table 3*). In addition, a blunted increase of HR contributed to the inappropriate response in CO, indicating that chronotropic incompetence is also involved in this group (PEF).

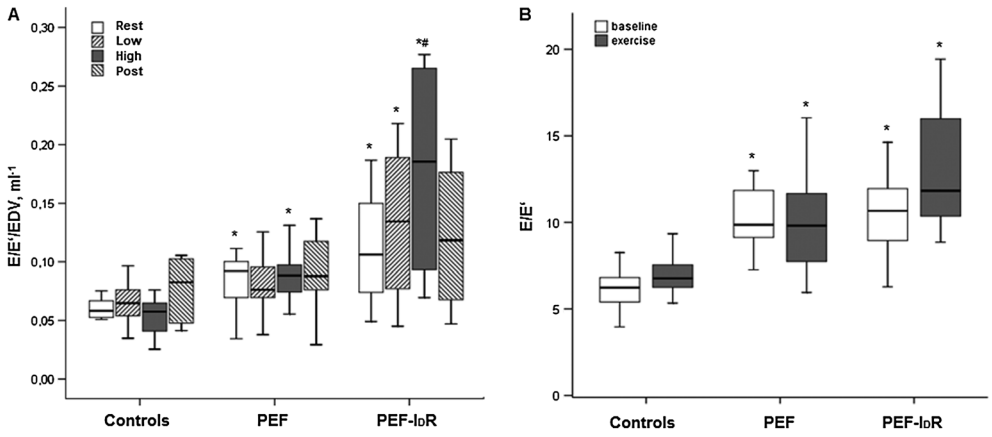
In the remaining 27 patients (52%), there was a continuous increase in *E*/*E*′ and (*E*/*E*′)/EDV during exercise (11.2 ± 3.7 to 16.8 ± 10.5 and 0.122 ± 0.038 to 0.217 ± 0.140/mL, respectively), suggesting impaired end-diastolic reserve (PEF-I_DR). This was associated with a fall in SV leading to a non-adequate increase in CO. The change of (*E*/*E*′)/EDV ratio during stress did not exceed 0.021/mL in PEF patients, and the increase of LV filling index remained under the cut-off value of 3.3 (*E*/*E*′ < 3.3, defined as the 90th percentile from the control group).

Table 3 Conventional and tissue Doppler imaging echocardiography during exercise in heart failure with preserved ejection fraction without *E/E'* increase (PEF) and with *E/E'* increase (PEF-I_DR) vs. controls (variable expressed as mean ± standard deviation)

		Controls ($n = 26$)	PEF ($n = 25$)	PEF-I_DR ($n = 27$)
Heart dimensions				
LA parasternal, mm		34 ± 4	37 ± 6	38 ± 8[a]
LAVI, mL/m^2		19 ± 12	21 ± 10	31 ± 17[ab]
Septum, mm		10 ± 1.5	12 ± 2.3	12 ± 1.8[a]
Posterior wall, mm		10 ± 1.5	11 ± 1.7	11 ± 1.5
LVEDVI, mL/m^2		57 ± 12	57 ± 15	55 ± 15
LVMI, g/m^2		97 ± 20	106 ± 34	119 ± 26[a]
Mitral flow				
E, cm/s	Baseline	74 ± 15	72 ± 12	78 ± 17
	Exercise	126 ± 25[c]	111 ± 29[c]	129 ± 22[c]
A, cm/s	Baseline	62 ± 17	73 ± 19[a]	81 ± 15[a]
	Exercise	97 ± 21[c]	95 ± 23[c]	117 ± 15[ac]
E/A	Baseline	1.25 ± 0.39	1.04 ± 0.32	0.98 ± 0.25
	Exercise	1.33 ± 0.41	1.10 ± 0.27	1.09 ± 0.20
Tissue Doppler				
S'_{mean}, cm/s	Baseline	8.4 ± 1.7	7.9 ± 2.2	7.6 ± 2.1
	Exercise	12.6 ± 2.5[c]	8.9 ± 2.8[ac]	8.8 ± 2.5[ac]
E'_{mean}, cm/s	Baseline	13.1 ± 3.1	7.3 ± 1.9[a]	7.5 ± 2.4[a]
	Exercise	19.2 ± 5.5[c]	11.7 ± 4.1[ac]	9.5 ± 3.8[ab]
A'_{mean}, cm/s	Baseline	8.5 ± 2.1	7.7 ± 2.4	8.5 ± 2.1
	Exercise	13.6 ± 4.5[c]	10.0 ± 3.1[c]	11.3 ± 3.9[c]
E' / A'_{mean}	Baseline	1.64 ± 0.52	1.09 ± 0.55[a]	0.93 ± 0.35[a]
	Exercise	1.34 ± 0.31	1.17 ± 0.45[ac]	0.89 ± 0.48[ab]
E' / E'_{mean}	Baseline	5.9 ± 1.2	10.5 ± 2.4[a]	11.2 ± 3.7[a]
	Exercise	6.9 ± 1.3	9.9 ± 2.7	16.8 ± 10.5[abc]
Speckle tracking				
2D long strain	Baseline	−21.2 ± 2.5	−19.4 ± 2.3	−19.2 ± 2.7
	Exercise	−24.8 ± 3.8[c]	−21.6 ± 1.2[c]	−22.4 ± 3.1[c]

2D, two dimensional; *E/A*, the ratio of early (*E*) to late (*A*) mitral flow peak velocities; *E'/A'* ratio of early to late annular velocity; *E/E'*, LV filling index; LA, left atrial; LAVI, left atrial volume index; LVEDVI, LV end-diastolic volume index; LVMI, LV mass index; *S'*, *E'*, and *A'*, systolic, early, and late diastolic peak velocities of mitral annulus at lateral site, respectively.
[a]$P < 0.05$ vs. controls; [b]$P < 0.05$ vs. PEF; [c]$P < 0.05$ baseline vs. exercise.

Figure 1 End-diastolic pressure–volume relationship (A) and *E/E'* (B) at baseline, during low and maximal exercise levels and recovery in heart failure with preserved ejection fraction without (PEF, $n = 25$) and with (PEF-I_DR, $n = 27$) *E/E'* increase vs. controls ($n = 26$). *$P < 0.05$ vs. controls; #$P < 0.05$ vs. baseline.



Between the two HFpEF subgroups, there was no difference in age, gender, or BMI. Among PEF patients, there was a tendency towards increased rates of diabetes mellitus (7/25 vs. 3/27, $P = 0.239$) and hyperlipoproteinaemia (16/25 vs. 7/27, $P = 0.056$) compared with PEF-I_DR. Both PEF and PEF-I_DR were characterized by exercise intolerance (NYHA classes II–III: 23/25 vs. 25/27, $P = 0.561$; exercise test: 107 ± 36 vs. 96 ± 34 W, $P = 0.175$), but NT-proBNP levels, which were very variable, were not significantly elevated in PEF-I_DR (271 ± 302 vs. 409 ± 904 pg/mL, $P = 0.498$). There

was also no difference in the utilization rate of heart failure medications, particularly beta-blockers (13/25 vs. 9/27, $P = 0.162$) and diuretics (12/25 vs. 11/27, $P = 0.532$) between subgroups.

Left ventricular volume changes and cardiac performance during exercise

The changes of LVEDV, ESV, SV, and EF during low and maximal exercise are shown in *Figure 2*. The controls responded with an initial increase in LVEDV (+16%) whereas left ventricular end-systolic volume (LVESV) remained unchanged, resulting in increased SV (+25%). At maximal exercise, LVEDV did not increase further, but LVESV decreased significantly (−27%). Despite only a mild additional increase in SV (+5%), CO increased significantly owing to an adequate chronotropic response (*Figure 2* and *Table 4*). In contrast, patients with increased (E/E')/EDV at exercise (PEF-I_DR) could not expand their LVEDV (−8%), and despite a decrease in LVESV (−16%), they showed decreased SV (−6%) at low-level exercise. At maximal exercise, they showed further decreases in LVEDV (−10%) and SV (−9%), which resulted in minimal increase in CO (ΔCO: 1.47 ± 1.25 vs. 8.2 ± 4.6 L/min, $P < 0.001$). Maximal SV and maximal CO were significantly lower in PEF-I_DR (*Table 4*), which was associated with a reduced exercise capacity (96 ± 34 vs. 178 ± 61 W, $P < 0.05$) and elevated NT-proBNP levels (409 ± 904 271 ± 302 vs. 65 ± 45 pg/mL, $P < 0.05$).

In PEF patients, E/E' and (E/E')/EDV did not increase further during exercise and showed cardiac performance indexes similar to controls with increases in LVEDV and SV (*Figure 2*). However, in comparison with controls, their increase in CO at exercise was significantly lower (ΔCO: 4.3 ± 3.1 vs. 8.2 ± 4.6 L/min, $P = 0.002$), resulting in a significantly lower maximal CO (*Table 4*).

(E/E')/EDV during exercise correlated inversely with LVEDV ($r = -0.67$, $P < 0.001$), SV ($r = -0.67$, $P < 0.001$; *Figure 3*), CO ($r = -0.62$, $P < 0.001$), and exercise level ($r = -0.40$, $P = 0.002$, *Figure 4*). (E/E')/EDV did not correlate with systolic indices such as LVESV, EF, or estimated LV ESP. The peak systolic annular velocity S' and longitudinal 2D strain did not differ among the groups at baseline or during exercise (*Table 3*) and showed no correlation with (E/E')/EDV.

Figure 2 Changes of end-diastolic volume, end-systolic volume, stroke volume, and ejection fraction during exercise in PEF ($n = 25$) and PEF-I_DR ($n = 27$) vs. controls ($n = 26$) according to the full-volume analysis by three-dimensional echocardiography. Absent expansion of left ventricular end-diastolic volume in PEF-I_DR was associated with lower stroke volume during exercise, whereas ejection fraction did not change during exercise in all groups. $P < 0.05$.

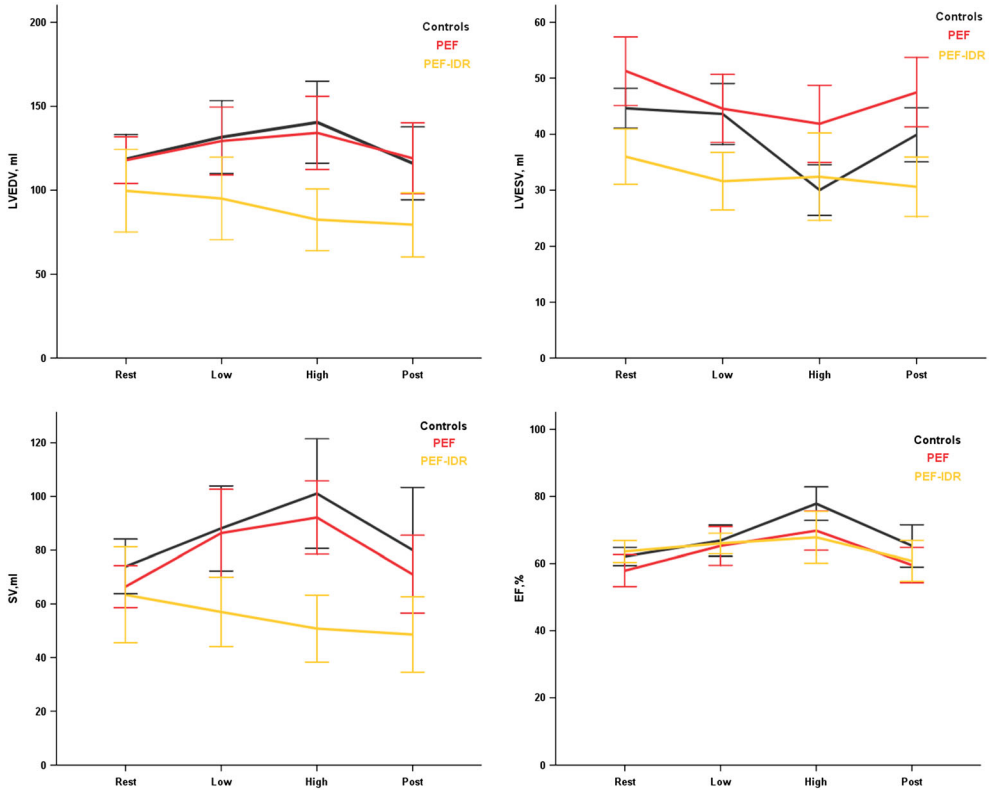


Table 4 Exercise and cardiac performance and systolic and diastolic function during bicycle exercise in heart failure with preserved ejection fraction vs. controls (variable expressed as mean ± standard deviation)

		Controls ($n = 26$)	PEF ($n = 25$)	PEF-I_DR ($n = 27$)
Exercise performance				
Maximal workload, W		178 ± 61	107 ± 36[a]	96 ± 34[a]
HR/min (% norm)	Baseline	77 ± 13	79 ± 13	80 ± 14
	Exercise	146 ± 20 (90 ± 9)	119 ± 21[a] (77 ± 17)[a]	134 ± 17[b](88 ± 5)
BP systolic, mmHg	Baseline	123 ± 19	136 ± 22[a]	147 ± 22[a]
	Exercise	178 ± 29	179 ± 28	185 ± 36
BP diastolic, mmHg	Baseline	71 ± 9	78 ± 13	74 ± 14
	Exercise	84 ± 13	87 ± 13	83 ± 14
Cardiac performance				
SV, mL	Baseline	68 ± 18	64 ± 19	63 ± 23[a]
	Exercise	92 ± 36[c]	73 ± 31	54 ± 24[ab]
CO, L/min	Baseline	5.3 ± 1.1	5.0 ± 1.9	5.2 ± 1.7
	Exercise	12.9 ± 4.4[c]	8.5 ± 3.5[a]	6.9 ± 3.0[ab]
Systolic indices				
EF, %	Baseline	62 ± 5	60 ± 8	63 ± 6
	Exercise	75 ± 9[c]	62 ± 11[ac]	64 ± 8[a]
LVESV, mL	Baseline	41 ± 13	47 ± 20	37 ± 12
	Exercise	33 ± 19[c]	44 ± 20[c]	30 ± 12[c]
eLVESP, mmHg	Baseline	116 ± 16	122 ± 20	132 ± 20
	Exercise	163 ± 31[c]	163 ± 31[c]	179 ± 24[c]
eEa, mmHg/mL	Baseline	1.7 ± 0.5	2.1 ± 0.6[a]	2.3 ± 0.7[a]
	Exercise	2.2 ± 0.6[c]	3.0 ± 0.8[ac]	3.0 ± 0.6[c]
eE_{LV}, mHg/mL	Baseline	3.0 ± 0.9	3.0 ± 1.3	3.7 ± 1.1
	Exercise	6.7 ± 3.4[c]	4.1 ± 1.5[c]	6.3 ± 1.9[c]
eEa/E_{LV}	Baseline	0.59 ± 0.12	0.74 ± 0.24	0.64 ± 0.24
	Exercise	0.38 ± 0.15[c]	0.65 ± 0.25	0.58 ± 0.21
Diastolic indices				
LVEDV, mL	Baseline	110 ± 29	111 ± 36	99 ± 32
	Exercise	129 ± 51	116 ± 45	83 ± 31[a]
E' / E'_{mean}	Baseline	5.9 ± 1.2	10.5 ± 2.4[a]	11.2 ± 3.7[a]
	Exercise	6.9 ± 1.3	9.9 ± 2.7	16.8 ± 10.5[abc]
E/E'/EDV/mL	Baseline	0.057 ± 0.016	0.099 ± 0.028[a]	0.122 ± 0.038[a]
	Exercise	0.061 ± 0.018	0.087 ± 0.031[a]	0.217 ± 0.14[acb]

BP, blood pressure; CO, cardiac output; Ea, arterial stiffness; Ea/E_{LV}, arterial–ventricular coupling; (E/E')/EDV, estimated end-diastolic pressure–volume relationship; EF, ejection fraction; EDV, end-diastolic volume; E_{LV}, LV end-systolic elastance; ESP, end-systolic pressure; ESV, end-systolic volume; HR, heart rate; LVESV, LV end-systolic volume; SV, stoke volume; SW, stroke work.
[a]$P < 0.05$ vs. controls;[b]$P < 0.05$ vs. PEF;[c]$P < 0.05$ baseline vs. exercise.

However, the increments of S' and 2D strain were more blunted in the PEF group showing a tendency to be different. According to multivariate regression analysis, (E/E')/EDV was found to be an independent predictor for cardiac performance in HFpEF patients (*Table 5*.)

Full-volume 3DE analysis showed low interobserver variability of EDV (1.6 ± 9.1 and 1.8 ± 10.3 mL), ESV (1.4 ± 7.6 and 2.0 ± 7.9 mL), and EF (0.2 ± 1.8% and 0.5 ± 4.3%, respectively) at rest and at exercise. Intraobserver variation was also low for EDV (−0.9 ± 7.3 and 1.2 ± 8.1 mL), ESV (−0.8 ± 5.6 and 1.0 ± 5.3 mL), and EF (0.1 ± 1.7% and 0.3 ± 2.9%, respectively).

Dyssynchrony—influence on left ventricular diastolic function and cardiac performance

According to the full-volume 3D analysis of the 16-segment regional volume changes, there were no differences in SDI between the groups at rest. Low time resolution of SDI did not allow assessment of this parameter during exercise because of the high HR. Only 12 HFpEF patients showed prolonged SDI at rest above the cut-off value of 40 ms, suggesting the presence of intraventricular mechanical dyssynchrony in these patients.[24,32] There were no significant correlations between SDI at rest and diastolic Doppler indices, SV, CO at exercise, NYHA class, or exercise level in this study group.

Arterial–ventricular coupling during exercise in heart failure with preserved ejection fraction

Estimated arterial elastance (eEa) was increased in the HFpEF group compared with controls (*Table 4*). At rest, HFpEF patients showed a tendency towards increased estimated end-systolic LV elastance (eE_{LV}) and smaller increases in eE_{LV} during exercise. Accordingly, arterial–ventricular coupling (eEa/eE_{LV}) only tended to be increased in HFpEF patients

Figure 3 Correlation of baseline and exercise *E*/*E*′/EDV with cardiac performance. Maximal stroke volume is related inversely to the increasing (A) baseline *E*/*E*′/EDV, SVmax = 15.3 + 4.1/(*E*/*E*′/EDV), *R* = −0.65, *P* < 0.001; and (B) exercise *E*/*E*′/EDV, SVmax = 31.9 + 3.2/(*E*/*E*′/EDV), *R* = −0.76, *P* < 0.001.

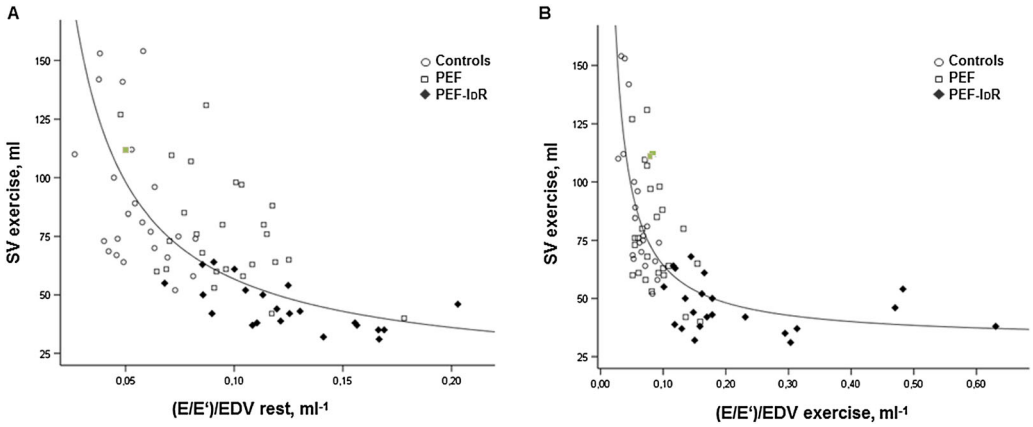


Table 5 Multivariate regression correlation coefficients (beta) with stroke volume at exercise and exercise capacity

	Stroke volume at stress		Exercise capacity (W)	
	Beta	*P*	Beta	*P*
E'_{mean}	0.51	0.001	0.76	0.001
E' / E'_{mean}	−1.10	0.001	−0.54	0.017
E/*E*′/EDV	−0.92	0.001	−0.45	0.021
Ees/Ea	0.29	0.053	0.12	0.456
SDI_{rest}	−0.31	0.755	−0.09	0.931
Chronotropy	−0.23	0.036	0.06	0.451

EDV, end-diastolic volume; Ees/Ea, ratio of end-systolic ventricular and arterial stiffening; SDI, systolic dyssynchrony index at rest.

and, perhaps more importantly, showed no significant decrease during exercise as was also exhibited in control patients (*Table 4*).

Discussion

We examined cardiac performance at rest and during exercise in controls, and in patients with HFpEF and borderline echocardiographic diastolic abnormalities at rest, using continuous bicycle echocardiography, 3D full-volume analysis and TDI. During exercise, controls showed the expected increase in LV contractility (eE_{LV}) and stable diastolic function as indicated by unchanged *E*/*E*′ values. In contrast, in addition to abnormal LV stiffness at rest and increased NT-proBNP values, all HFpEF patients had decreased exercise capacities with inability to increase adequately SV and CO, and they had abnormal LV–arterial coupling. However, there were different responses to exercise among HFpEF patients such that two distinctly different haemodynamic patterns emerged. First, there was a group of HFpEF patients who failed to increase SV who had a blunted increase in eE_{LV} but who showed no changes in *E*/*E*′. Despite observed abnormalities, particularly in early diastolic function (*E*′), an impaired systolic reserve and chronotropic incompetence appeared to primarily underlie exercise-induced symptoms in the majority of this group (designated PEF). Thus, several cardiac function disturbances were observed in this group, including also chronotropic incompetence indicating a heterogeneous subgroup that may as well involve early, not fully manifested, stages of the disease. In contrast, the second group showed near-normal increases in LV contractility but was characterized by a marked increase of *E*/*E*′, suggestive of increased filling pressure and associated reduction of LV compliance. Therefore, impaired diastolic reserve (PEF-I_DR) appeared to contribute to exercise-induced symptoms in this group. Interestingly, the rise in *E*/*E*′ was driven by a failure to augment *E*′ in early diastole, which is preload dependent. During exercise, early diastolic movement (*E*′) more sensitively reflects long-axis diastolic abnormalities than changes in global diastolic function (mitral early diastolic flow, *E*). *E*′ together with *E*/*E*′ belongs to important measurements for the evaluation of diastole in HFpEF. In summary, these data underscore the need to extend current guidelines for the diagnosis of HFpEF to include physiological investigations during exercise.

The pathophysiological mechanisms responsible for exercise limitations in HFpEF have not been fully clarified despite several recent studies. Inability to adequately increase CO and maintain low pulmonary capillary wedge pressures are common themes. Suggested underlying mechanisms include abnormal contractile reserve, worsening of diastolic dysfunction with impaired relaxation and increased LV stiffness,

impaired arterial–ventricular coupling, and mechanical intraventricular dyssynchrony and reduced chronotropic reserve.[3,16,17,21,33–37]

Determination of whether diastolic dysfunction plays a primary role in limiting exercise capacity in HFpEF patients is a complex matter, in particular when resting values from Doppler and TDI reveal borderline results in patients without a long history of typical risk factors, LV hypertrophy, or LA dilatation.[38] TDI-based parameters of diastole measured at rest were found to be the strongest echocardiographic predictors of exercise intolerance in HFpEF.[39] However, we suggest that TDI together with 3D echocardiography allows for estimation of end-diastolic pressure in addition to providing non-invasive assessment of pressure–volume-based assessment of diastolic stiffness, both at rest and during exercise.

Importantly, we investigated patients with HFpEF diagnosed on the basis of objective tests, including ergometry and NT-proBNP levels, who had borderline resting *E*/*E*′ values but who did not meet current TDI diagnostic criteria for HFpEF.

Among our cohort of patients with such borderline resting diastolic dysfunction, 52% showed further and significant increases in *E*/*E*′ during exercise (suggesting marked increases in LV end-diastolic pressure), which was associated with increased stiffening as indexed by increased *E*/*E*′/EDV during exercise (*Figure 1*). Such additional LV stiffening during exercise was observed before[40] and may result from an exercise-induced myocardial energy deficit and paradoxical prolongation of LV relaxation.[41] These observations are consistent with the hypothesis that the hearts of these PEF-I_DR patients are working within a steep portion of the ventricular diastolic compliance curve, which markedly limits the ability to increase SV. As a result, the increase in CO during exercise (normally due to increases in SV and HR) was markedly blunted compared with that observed in the control group and even in PEF patients. This is synonymous with the concept that these PEF-I_DR patients are working at high filling pressures during exercise, near the plateau of their Starling curves where further increases in filling pressure do not increase SV.[15,35,42,43] This is also in line with our finding that LA size was significantly enlarged in PEF-I_DR compared with controls and PEF. In comparison, LV elastance of PEF-I_DR did not differ from controls at rest or during exercise, suggesting that the impairment of diastolic reserve was the main mechanism contributing to exercise intolerance in this HFpEF subpopulation. Our non-invasive data further demonstrate that increased filling pressures occur in those HFpEF patients without overt total body volume overload or chronic enlargement of the LV.

However, these findings do not translate to all of HFpEF, even in our small cohort of patients. The PEF subgroup showed no pathological increase in *E*/*E*′/EDV during exercise (*Figure 1*), suggesting preservation of LV diastolic compliance as was seen in the control group. In contrast, it was an inability to adequately increase LV early diastolic filling and contractile function that contributed to blunted increases in SV and CO (*Table 4*). Although we could not clarify the underlying mechanism leading to impaired systolic reserve, we suggest that impaired chronotropic response could be a fundamental contributing factor (*Table 4*). Such chronotropic incompetence was observed independent of medication use; in particular, the frequency of beta-blocker use was similar between the two HFpEF subgroups.

While the identification of distinct phenotypic HFpEF subgroups could reflect patients with fundamentally different underlying diseases, we cannot exclude that these reflect different stages of the same disease and whether PEF patients will evolve into the PEF-I_DR phenotype over time. There were no differences in basic demographic characteristics between groups that would suggest this to be the case. In addition, there is ongoing debate whether non-diastolic (and even non-cardiac) aspects of cardiovascular function may dominate as the cause of limiting exercise capacity in some HFpEF patients. Proposed mechanisms include LV dyssynchrony,[44,45] reduced LV contractility,[14,17,35,46–48] abnormal arterial–ventricular coupling,[16,49] and chronotropic incompetence.[3,50] We did identify degrees of abnormal arterial–ventricular coupling, lack of appropriate vasodilation leading to more unfavourable arterial–ventricular coupling, and decreased LV contractility reserve in our study population. Proposed non-cardiac mechanisms include abnormal salt and water handling due to renal dysfunction and abnormal autonomic regulation of venous tone at rest and during exercise.[5,47] Thus, results from prior studies and data from our own study demonstrate that different mechanisms may contribute to exercise intolerance and that different patients may manifest exercise intolerance as a consequence of different underlying mechanisms. For a given patient, this may depend on the nature and severity of risk factors and co-morbid conditions, including ageing, the duration and severity of hypertension, the presence of prior infarctions or coronary artery disease, renal dysfunction, and arrhythmias such as atrial fibrillation. In our overall study population where coronary artery disease and atrial fibrillation were excluded, the PEF subgroup accounted for 48% of the entire cohort; the evidence suggests that in these subjects, non-diastolic mechanisms (such as chronotropic incompetence and/or reduced systolic reserve) dominated.

Limitations

Patients with atrial fibrillation and severe coronary artery disease were excluded in the study in order to remove these as potential confounding factors in the analysis. However, the inclusion criteria and baseline characteristics may define a

particular subset of patients and might account for some differences from other studies.

Some caution is needed when interpreting 3DE data acquired during higher HRs because of limited temporal resolution. According to the recommendations, 20 fps are needed for digital capture at normal HRs, and frame rates should ideally be increased to 30 fps when HR is over 140 bpm.[51] Accordingly, 3DE acquisition was performed at the high frame rate at an exercise level >65% maximal workload, which also assured HRs lower than 140 bpm. After overcoming a learning curve and using multibeat breath holds, it is possible to acquire volume data under exercise condition with high reliability.[52]

Another limitation is that more detailed although cumbersome approaches are available for non-invasive quantification of diastolic and systolic ventricular properties through estimated pressure–volume analysis.[53,54] We performed an analysis using these techniques, and the conclusions are not altered. Thus, the simple approaches used here appear to capture the essence of differences in systolic and diastolic properties between groups.

Conclusions

In summary, our data show that 3DE volume analysis provides accurate measurements of volumes that can be used to quantify diastolic and systolic LV functions in HFpEF at rest and during exercise. A combination of TDI measurement of *E*/*E*′ along with EDV to arrive at (*E*/*E*′)/EDV, a non-invasive index of diastolic stiffness, can help to determine the degree to which diastolic mechanisms contribute to reduced cardiac reserve and exercise intolerance. Similarly, the ratio between estimated ESP and ESV provides an index of LV contractility that can be tracked at rest and during exercise to help elucidate the role of contractile reserve and arterial–ventricular coupling.

Consistent with the growing literature on exercise intolerance in HFpEF, we observed that different diastolic and non-diastolic mechanisms can be identified during exercise in HFpEF and that exercise tolerance can be limited in different patients to varying degrees by those different mechanisms. 3D echocardiography may be used to discriminate such abnormalities that may help to clarify pathophysiologic mechanisms in the heterogeneous pathophysiology of HFpEF. This has a particular impact when patients with borderline findings at rest need to be further evaluated by exercise testing. Additional population-based studies need to evaluate whether these findings will contribute to the understanding of prognosis and treatment options in HFpEF.

Acknowledgements

M. K. was a recipient of the European Association of Echocardiography research grant 2008. This study was also supported by the EC, FP7-Health-2010, MEDIA (261409) to C. T. and A. G. F. We thank Conny Lober for the great support in performing exercise tests.

Disclosures

D. Burkhoff receives support from DC Devices to run a haemodynamic core lab for a study investigating a device-based therapy for patients with HFpEF.

References

1. Owan TE, Hodge DO, Herges RM, Jacobsen SJ, Roger VL, Redfield MM. Trends in prevalence and outcome of heart failure with preserved ejection fraction. *N Engl J Med* 2006; **355**: 251–259.
2. Bhatia RS, Tu JV, Lee DS, Austin PC, Fang J, Haouzi A, Gong Y, Liu PP. Outcome of heart failure with preserved ejection fraction in a population-based study. *N Engl J Med* 2006; **355**: 260–269.
3. Borlaug BA, Melenovsky V, Russell SD, Kessler K, Pacak K, Becker LC, Kass DA. Impaired chronotropic and vasodilator reserves limit exercise capacity in patients with heart failure and a preserved ejection fraction. *Circulation* 2006; **114**: 2138–2147.
4. Kawaguchi M, Hay I, Fetics B, Kass DA. Combined ventricular systolic and arterial stiffening in patients with heart failure and preserved ejection fraction: implications for systolic and diastolic reserve limitations. *Circulation* 2003; **107**: 714–720.
5. Maurer MS, Burkhoff D, Fried LP, Gottdiener J, King DL, Kitzman DW. Ventricular structure and function in hypertensive participants with heart failure and a normal ejection fraction: the Cardiovascular Health Study. *J Am Coll Cardiol* 2007; **49**: 972–981.
6. Kasner M, Westermann D, Steendijk P, Drose S, Poller W, Schultheiss HP, Tschope C. Left ventricular dysfunction induced by nonsevere idiopathic pulmonary arterial hypertension: a pressure–volume relationship study. *Am J Respir Crit Care Med* 2012; **186**: 181–189.
7. Borbely A, van der Velden J, Papp Z, Bronzwaer JG, Edes I, Stienen GJ, Paulus WJ. Cardiomyocyte stiffness in diastolic heart failure. *Circulation* 2005; **111**: 774–781.
8. Katz AM, Zile MR. New molecular mechanism in diastolic heart failure. *Circulation* 2006; **113**: 1922–1925.
9. Querejeta R, Lopez B, Gonzalez A, Sanchez E, Larman M, Martinez Ubago JL, Diez J. Increased collagen type I synthesis in patients with heart failure of hypertensive origin: relation to myocardial fibrosis. *Circulation* 2004; **110**: 1263–1268.
10. De Keulenaer GW, Brutsaert DL. Diastolic heart failure: a separate disease or selection bias? *Prog Cardiovasc Dis* 2007; **49**: 275–283.
11. Senni M, Paulus WJ, Gavazzi A, Fraser AG, Diez J, Solomon SD, Smiseth OA, Guazzi M, Lam CS, Maggioni AP, Tschope C, Metra M, Hummel SL, Edelmann F, Ambrosio G,

Stewart Coats AJ, Filippatos GS, Gheorghiade M, Anker SD, Levy D, Pfeffer MA, Stough WG, Pieske BM. New strategies for heart failure with preserved ejection fraction: the importance of targeted therapies for heart failure phenotypes. *Eur Heart J* 2014; **35**: 2797–2815.
12. Tschope C, Van Linthout S. New insights in (inter)cellular mechanisms by heart failure with preserved ejection fraction. *Current Heart Failure Reports* 2014.
13. Kasner M, Westermann D, Lopez B, Gaub R, Escher F, Kuhl U, Schultheiss HP, Tschope C. Diastolic tissue Doppler indexes correlate with the degree of collagen expression and cross-linking in heart failure and normal ejection fraction. *J Am Coll Cardiol* 2011; **57**: 977–985.
14. Kasner M, Westermann D, Steendijk P, Gaub R, Wilkenshoff U, Weitmann K, Hoffmann W, Poller W, Schultheiss HP, Pauschinger M, Tschope C. Utility of Doppler echocardiography and tissue Doppler imaging in the estimation of diastolic function in heart failure with normal ejection fraction: a comparative Doppler-conductance catheterization study. *Circulation* 2007; **116**: 637–647.
15. Zile MR, Baicu CF, Gaasch WH. Diastolic heart failure—abnormalities in active relaxation and passive stiffness of the left ventricle. *N Engl J Med* 2004; **350**: 1953–1959.
16. Lam CS, Roger VL, Rodeheffer RJ, Bursi F, Borlaug BA, Ommen SR, Kass DA, Redfield MM. Cardiac structure and ventricular–vascular function in persons with heart failure and preserved ejection fraction from Olmsted County, Minnesota. *Circulation* 2007; **115**: 1982–1990.
17. Redfield MM, Jacobsen SJ, Borlaug BA, Rodeheffer RJ, Kass DA. Age- and gender-related ventricular–vascular stiffening: a community-based study. *Circulation* 2005; **112**: 2254–2262.
18. Maurer MS, Mancini D. HFpEF: is splitting into distinct phenotypes by comorbidities the pathway forward? *J Am Coll Cardiol* 2014; **64**: 550–552.
19. Shah SJ, Katz DH, Selvaraj S, Burke MA, Yancy CW, Gheorghiade M, Bonow RO, Huang CC, Deo RC. Phenomapping for novel classification of heart failure with preserved ejection fraction. *Circulation* 2015; **131**: 269–279.
20. Paulus WJ, Tschope C, Sanderson JE, Rusconi C, Flachskampf FA, Rademakers FE, Marino P, Smiseth OA, De Keulenaer G, Leite-Moreira AF, Borbely A, Edes I, Handoko ML, Heymans S, Pezzali N, Pieske B, Dickstein K, Fraser AG, Brutsaert DL. How to diagnose diastolic heart failure: a consensus statement on the diagnosis of heart failure with normal left ventricular ejection fraction by the Heart Failure and Echocardiography Associations of the European Society of Cardiology. *Eur Heart J* 2007; **28**: 2539–2550.
21. Borlaug BA. Mechanisms of exercise intolerance in heart failure with preserved ejection fraction. *Circ J: Offic J Jpn Circ Soc* 2014; **78**: 20–32.
22. Tschope C, Kasner M, Westermann D, Walther T, Gaub R, Poller WC, Schultheiss HP. Elevated NT-ProBNP levels in patients with increased left ventricular filling pressure during exercise despite preserved systolic function. *J Card Fail* 2005; **11**: S28–S33.
23. Fletcher GF, Balady GJ, Amsterdam EA, Chaitman B, Eckel R, Fleg J, Froelicher VF, Leon AS, Pina IL, Rodney R, Simons-Morton DA, Williams MA, Bazzarre T. Exercise standards for testing and training: a statement for healthcare professionals from the American Heart Association. *Circulation* 2001; **104**: 1694–1740.
24. Kapetanakis S, Kearney MT, Siva A, Gall N, Cooklin M, Monaghan MJ. Real-time three-dimensional echocardiography: a novel technique to quantify global left ventricular mechanical dyssynchrony. *Circulation* 2005; **112**: 992–1000.
25. Kasner M, Sinning D, Burkhoff D, Tschope C. Diastolic pressure–volume quotient (DPVQ) as a novel echocardiographic index for estimation of LV stiffness in HFpEF. *Clin Res Cardiol* 2015. [Epub ahead of print] doi: 10.1007/s00392-015-0863-y.
26. Kelly RP, Ting CT, Yang TM, Liu CP, Maughan WL, Chang MS, Kass DA. Effective arterial elastance as index of arterial vascular load in humans. *Circulation* 1992; **86**: 513–521.
27. Burkhoff D, Mirsky I, Suga H. Assessment of systolic and diastolic ventricular properties via pressure–volume analysis: a guide for clinical, translational, and basic researchers. *Am J Physiol* 2005; **289**: H501–H512.
28. Kass DA, Kelly RP. Ventriculo-arterial coupling: concepts, assumptions, and applications. *Ann Biomed Eng* 1992; **20**: 41–62.
29. Sunagawa K, Maughan WL, Burkhoff D, Sagawa K. Left ventricular interaction with arterial load studied in isolated canine ventricle. *Am J Physiol* 1983; **245**: H773–H780.
30. Reisner SA, Lysyansky P, Agmon Y, Mutlak D, Lessick J, Friedman Z. Global longitudinal strain: a novel index of left ventricular systolic function. *J Am Soc Echocardiogr* 2004; **17**: 630–633.
31. Rossi A, Cicoira M, Florea VG, Golia G, Florea ND, Khan AA, Murray ST, Nguyen JT, O'Callaghan P, Anand IS, Coats A, Zardini P, Vassanelli C, Henein M. Chronic heart failure with preserved left ventricular ejection fraction: diagnostic and prognostic value of left atrial size. *Int J Cardiol* 2006; **110**: 386–392.
32. Kasner M, Westermann D, Schultheiss HP, Tschope C. Diastolic heart failure and LV dyssynchrony. *Curr Pharm Biotechnol* 2012; **13**: 2539–2544.
33. Borlaug BA, Paulus WJ. Heart failure with preserved ejection fraction: pathophysiology, diagnosis, and treatment. *Eur Heart J* 2011; **32**: 670–679.
34. Kraigher-Krainer E, Shah AM, Gupta DK, Santos A, Claggett B, Pieske B, Zile MR, Voors AA, Lefkowitz MP, Packer M, McMurray JJ, Solomon SD, Investigators P. Impaired systolic function by strain imaging in heart failure with preserved ejection fraction. *J Am Coll Cardiol* 2014; **63**: 447–456.
35. Westermann D, Kasner M, Steendijk P, Spillmann F, Riad A, Weitmann K, Hoffmann W, Poller W, Pauschinger M, Schultheiss HP, Tschope C. Role of left ventricular stiffness in heart failure with normal ejection fraction. *Circulation* 2008; **117**: 2051–2060.
36. Sinning D, Kasner M, Westermann D, Schulze K, Schultheiss HP and Tschope C. Increased left ventricular stiffness impairs exercise capacity in patients with heart failure symptoms despite normal left ventricular ejection fraction. *Cardiol Res Pract* 2011; **2011**: 1–10, 692862.
37. Kasner M, Gaub R, Sinning D, Westermann D, Steendijk P, Hoffmann W, Schultheiss HP, Tschope C. Global strain rate imaging for the estimation of diastolic function in HFNEF compared with pressure–volume loop analysis. *Eur J Echocardiogr* 2010; **11**: 743–751
38. Tschope C, Westermann D. Heart failure with normal ejection fraction. Pathophysiology, diagnosis, and treatment. *Herz* 2009; **34**: 89–96.
39. Grewal J, McCully RB, Kane GC, Lam C, Pellikka PA. Left ventricular function and exercise capacity. *Jama* 2009; **301**: 286–294.
40. Borlaug BA, Jaber WA, Ommen SR, Lam CS, Redfield MM, Nishimura RA. Diastolic relaxation and compliance reserve during dynamic exercise in heart failure with preserved ejection fraction. *Heart (Br Card Soc)* 2011; **97**: 964–969.
41. Phan TT, Abozguia K, Nallur Shivu G, Mahadevan G, Ahmed I, Williams L, Dwivedi G, Patel K, Steendijk P, Ashrafian H, Henning A, Frenneaux M. Heart failure with preserved ejection fraction is characterized by dynamic impairment of active relaxation and contraction of the left ventricle on exercise and associated with myocardial energy deficiency. *J Am Coll Cardiol* 2009; **54**: 402–409.
42. Kitzman DW, Higginbotham MB, Cobb FR, Sheikh KH, Sullivan MJ. Exercise intolerance in patients with heart failure and preserved left ventricular systolic function: failure of the Frank–Starling mechanism. *J Am Coll Cardiol* 1991; **17**: 1065–1072.
43. Wachter R, Schmidt-Schweda S, Westermann D, Post H, Edelmann F, Kasner M, Luers C, Steendijk P, Hasenfuss G, Tschope C, Pieske B. Blunted frequency-dependent upregulation of cardiac output is related to impaired relaxation in diastolic heart failure. *Eur Heart J* 2009; **30**: 3027–3036.
44. Wang J, Kurrelmeyer KM, Torre-Amione G, Nagueh SF. Systolic and diastolic dyssynchrony in patients with diastolic

heart failure and the effect of medical therapy. *J Am Coll Cardiol* 2007; **49**: 88–96.
45. Wang YC, Hwang JJ, Lai LP, Tsai CT, Lin LC, Katra R, Lin JL. Coexistence and exercise exacerbation of intraleft ventricular contractile dyssynchrony in hypertensive patients with diastolic heart failure. *Am Heart J* 2007; **154**: 278–284.
46. Baicu CF, Zile MR, Aurigemma GP, Gaasch WH. Left ventricular systolic performance, function, and contractility in patients with diastolic heart failure. *Circulation* 2005; **111**: 2306–2312.
47. Zile MR, Gaasch WH, Carroll JD, Feldman MD, Aurigemma GP, Schaer GL, Ghali JK, Liebson PR. Heart failure with a normal ejection fraction: is measurement of diastolic function necessary to make the diagnosis of diastolic heart failure? *Circulation* 2001; **104**: 779–782.
48. Tan YT, Wenzelburger F, Lee E, Heatlie G, Leyva F, Patel K, Frenneaux M, Sanderson JE. The pathophysiology of heart failure with normal ejection fraction: exercise echocardiography reveals complex abnormalities of both systolic and diastolic ventricular function involving torsion, untwist, and longitudinal motion. *J Am Coll Cardiol* 2009; **54**: 36–46.
49. Maurer MS, King DL, El-Khoury Rumbarger L, Packer M, Burkhoff D. Left heart failure with a normal ejection fraction: identification of different pathophysiologic mechanisms. *J Card Fail* 2005; **11**: 177–187.
50. Brubaker PH, Joo KC, Stewart KP, Fray B, Moore B, Kitzman DW. Chronotropic incompetence and its contribution to exercise intolerance in older heart failure patients. *J Cardiopulm Rehabil* 2006; **26**: 86–89.
51. Armstrong WF, Pellikka PA, Ryan T, Crouse L, Zoghbi WA. Stress echocardiography: recommendations for erformance and interpretation of stress echocardiography. Stress Echocardiography Task Force of the Nomenclature and Standards Committee of the American Society of Echocardiography. *J Am Soc Echocardiogr* 1998; **11**: 97–104.
52. Pratali L, Molinaro S, Corciu AI, Pasanisi EM, Scalese M, Sicari R. Feasibility of real-time three-dimensional stress echocardiography: pharmacological and semi-supine exercise. *Cardiovasc Ultrasound* 2010; **8**: 10–18.
53. Klotz S, Hay I, Dickstein ML, Yi GH, Wang J, Maurer MS, Kass DA, Burkhoff D. Single-beat estimation of end-diastolic pressure–volume relationship: a novel method with potential for noninvasive application. *Am J Physiol* 2006; **291**: H403–H412.
54. Senzaki H, Chen CH, Kass DA. Single-beat estimation of end-systolic pressure–volume relation in humans. A new method with the potential for noninvasive application. *Circulation* 1996; **94**: 2497–2506.